

COMPARATIVE ANALYSIS OF CALCIUM IN PHARMACEUTICAL AND NUTRACEUTICAL PRODUCT BY CLASSICAL AND NON-CLASSICAL METHODS

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ABSTRACT

This study investigates the calcium content in two pharmaceutical products, calcium lactate (300 mg) and calcium gluconate (500 mg), and two nutraceutical dietary supplements, milk powder and yogurt, through both qualitative and quantitative analysis. Qualitative analysis was performed using flame tests and visible spectrophotometry, with all samples showing a brick red flame color and a maximum absorbance wavelength of 520 nm, confirming the presence of calcium. Quantitative analysis was conducted using volumetric titration and UV-visible spectrophotometry. The results showed that the calcium content of calcium gluconate was 97.76% by UV spectroscopy and 96.41% by titration, while calcium lactate exhibited 100.33% and 98.64%, respectively. Milk powder and yogurt showed lower calcium contents of 12% and 18.1%, respectively in UV spectroscopy and with titration results of 9.33% and 15.2%. The comparison of classical methods (flame test and titration) with non-classical UV-visible spectrophotometry revealed that the latter is more efficient, offering a unified approach for both identification and quantification of calcium. The study concludes that visible spectrophotometric analysis provides a more streamlined and accurate means of assessing calcium content in pharmaceutical and nutraceutical products.

KEYWORDS: Calcium content, Pharmaceutical products, Nutraceutical dietary supplements, UV-visible spectrophotometry, Quantitative analysis.

INTRODUCTION

Pharmaceutical Analysis^[1]

Pharmaceutical analysis is the series of process that are used for identification, determination, separation, purification and structure elucidation of the given compound used in the formulation of pharmaceutical products. Used to determine the purity, safety and quality of drugs and chemicals. The term “Pharmaceutical Analysis” is otherwise called Quantitative pharmaceutical chemistry. Pharmaceutical analysis includes both qualitative and quantitative analysis of drugs and pharmaceutical substance starts from bulk drugs (starting materials) to the finished dosage form.

Qualitative Analysis

It may be defined as “which analyte present in the given sample”. This has the goal to identify the various components present in the given sample on the basis of physical and chemical properties such as functional groups elemental composition and melting point etc.

Quantitative Analysis

It may be defined as “how much amount of analyte present in the given sample”. This has the goal to determine the quantity of each component present in the given sample.

Applications

- ❖ Classification of a compound according to its chemical properties.
- ❖ Analysis of mixture for compounds.
- ❖ Separating components from mixtures.
- ❖ Purification, identification and characterization of compounds.

Classical Analysis

Classical analysis is the group of analytical methods that only requires the use of chemical, a balance, calibrated glassware and other common place laboratory apparatus, such as funnels, burners or hot plates, flasks and beakers.

Titration Method

Titration

Titration is the process in the standard solution (Normal/Molar solution) is added in a controlled manner from burette to the sample (or analyte) to be estimated until the reaction is just complete.

Non-Classical Analysis

Non-classical analysis requires the use of an analytical instrument in addition the apparatus that is used for classical analysis. Classical and non-classical analysis methods can be used for qualitative and quantitative analysis. An analytical instrument is a physical, often electrically operated, device that is used to determine the identify or amount of one or more components in the analyzed substance.

Spectroscopy^[2]

Introduction

Spectroscopy is the measurement and interpretation of electromagnetic radiation (EMR) absorbed or emitted when the molecule or atoms or ions of a sample move from one energy state to another energy state. This change may be from ground state to excited state or excited to ground state. At ground state, the energy of a molecule is the sum total of

rotational, vibrational and electronic energies. In other words, spectroscopy measure the changes in rotational, vibrational and/or electronic energies.

UV-Spectroscopy

UV-Vis Spectroscopy (or Spectrophotometry) is a quantitative technique used to measure how much a chemical substance absorbs light. This is done by measuring the intensity of light that passes through a sample with respect to the intensity of light through a reference sample or blank.

UV-Vis Spectroscopy is routinely used in analytical chemistry for the quantitative determination of different analytes or sample, such as transition metal ions, highly conjugated organic compounds, and biological macromolecules.

Principle

The Principle of UV-Visible Spectroscopy is based on the absorption of ultraviolet light or visible light by chemical compounds, which results in the production of distinct spectra.

When the matter absorbs the light, it undergoes excitation and de-excitation, resulting in the production of a spectrum.

Beer's Law

This law states that "the intensity of a beam of monochromatic light decreases exponentially with increase in the concentration of absorbing species arithmetically".

$$\frac{-dI}{dc} \propto I$$

Lambert's Law

The rate of decrease of intensity (monochromatic light) with the thickness of the medium is directly proportional to the intensity of incident light.

$$\frac{-dI}{dt} \propto I$$

Mathematical equation for beer-lambert's law,

$$A = \epsilon ct$$

METHODS OF UV-SPECTROSCOPY

- Simultaneous equation method.
- Derivative spectrophotometric method.
- Absorbance ratio method (Q-Absorbance method)
- $E_{1\text{cm}}^{1\%}$ Difference spectrophotometry Solvent extraction method.

$E_{1\text{cm}}^{1\%}$ means the absorbance of 1% w/v solution using a path length of 1cm.

$E_{1\text{cm}}^{1\%}$ at a wavelength is a constant value for each can be seen in pharmacopoeias and standard books on subject. This value is useful in determining the concentration of drug in sample formulations or in solutions. E_{max} is the value at λ_{max} .

Calcium

Calcium is the most abundant mineral in the body. Almost all calcium in the body is stored in bones and teeth, giving them structure and hardness. Your body needs calcium for muscles to move and for nerves to carry messages between your brain and every part of your body.

Calcium is now used to preserve various foodstuffs and prolong shelf life of food. Its main applications include preservation of vegetables, fruits, meat and meat products, also being added in beverages, jelly, chewing gum, candy products, calcium lactate can be used in food industry as calcium source, dietary supplement, for preserving fresh food as antioxidant and stabilizer, as antimicrobial. It can be produced by chemical reaction between lactic acid and calcium carbonate.

Calcium has antimicrobial activity against aerobic and anaerobic microorganisms that can be found in meat. Although calcium lactate, sodium lac.

What Are Synthetic and Natural Nutrients?

Natural nutrients

- ❖ These are obtained from whole food sources in the diet.

Synthetic nutrients

- ❖ Synthetic nutrients do not include “whole food supplements,” which are made from concentrated, dehydrated whole foods.
- ❖ The majority of supplements available on the market today are made artificially. These include vitamins, antioxidants, minerals and amino acids, among others.
- ❖ They can be taken in pill, capsule, tablet, powder or liquid form, and are made to mimic the way natural nutrients act in our bodies.^[3]

Calcium-Rich Foods

- People can obtain calcium from a range of foods and drinks.

The following are good sources:

- ❖ Milk and Yogurt.
- ❖ Fortified dairy alternatives, such as soy milk.
- ❖ Sardines and salmon.
- ❖ Cheese and tofu.
- ❖ Green leafy vegetables, such as broccoli, turnip leaves, watercress, and kale.
- ❖ Fortified fruit juices.
- ❖ Nuts and seeds, especially almonds, sesame, and chia.
- ❖ Cornmeal and corn tortillas.

Calcium Deficiency

The following conditions or lifestyle habits may result in low calcium levels, also known as hypocalcemia:

- Bulimia, anorexia, and some other eating disorders.
- Mercury exposure.

- Overconsumption of magnesium.
- Lack of parathyroid hormone.
- People who eat a lot of protein or sodium may excrete calcium.
- Some cancers.
- Some Conditions such as celiac disease and inflammatory disease, Crohn's disease, and some other digestive diseases.
- Some surgical procedures, including removing the stomach.
- Kidney failure.
- Vitamin D deficiency.
- Phosphate deficiency.

Possible complication

- Kidney stones.
- A reduction in iron absorption.
- A higher risk of a heart attack.
- However, more recent studies have suggested that these concerns may be unfounded.
- Calcium may interact with some drugs.^[5]

Pharmaceutical

Pharmaceutical, substance used in the diagnosis, treatment or prevention of disease and for restoring, correcting or modifying organic functions.

Pharmaceuticals are generally classified by chemical group, by the way they work in the body (pharmacological effect), and by therapeutic use.

Antibiotics, vaccines, human blood-plasma fractions, and steroid hormones are other important pharmaceuticals manufactured from natural substances.

Pharmaceutical product

Pharmaceutical products consist of active ingredients, which are combined with additional materials (excipients) selected to control dosage delivery, enhance performance and facilitate manufacture.

Pharmaceutical manufacturing

Pharmaceutical manufacturing traditionally uses batch operations and is generally carried out in multipurpose plants.

There is much benefit to be derived from the development and application of scheduling methodology, which would drive improvements in capital utilization and from the use of process control and dynamic optimization methodologies to drive reduction in batch-to-batch variability.

Calcium Lactate

Calcium lactate is a white crystalline salt with formula $C_6H_{10}CaO_6$, consisting of two lactate anions $H_3CCO_2^-$ for each calcium cation Ca^{2+} . It forms several hydrates, the most common being the pentahydrate $C_6H_{10}CaO_6 \cdot 5H_2O$.

- **Formula:** $C_6H_{10}CaO_6$

- **Molar mass:** 218.22 g/mol
- **Solubility:** Very soluble in methanol, insoluble in ethanol
- **Density:** 1.49 g/cm³
- **Acidity (pK_a):** 6.0-8.5
- **ATC code:** A12AA05 (WHO).^[6]

Definition

Calcium lactate is a food additive that's typically added to a wide variety of foods to enhance their texture and flavor or help extend their shelf life.

This compound can also be used as an ingredient in medications or certain types of calcium supplements.^[7]

Possible health benefits

- ❖ Stronger bones
- ❖ Reduced blood pressure
- ❖ Protection against preeclampsia
- ❖ Protection against colon cancer

Calcium Gluconate

Calcium gluconate is the calcium salt of gluconic acid and is used as a mineral supplement and medication. As a medication it is used by injection into a vein to treat low blood calcium, high blood potassium, and magnesium toxicity. Supplementation is generally only required when there is not enough calcium in the diet. Supplementation may be done to treat or prevent osteoporosis or rickets. It can also be taken by mouth but is not recommended for injection into a muscle.

ChEMBL Id: 2106119

ChemSpider ID: 8932

Formula: C₁₂H₂₂CaO₁₄

IUPAC ID: calcium (2R,3S,4R,5R)- 2,3,4,5,6-pentahydroxyhexanoate

Melting point: 120 °C

Molar mass: 430.373 g/mol

Soluble in: Water

NUTRACEUTICALS

Nutraceutical is a latest term for health food, first innovated by Stephen Deffice, founder of the Foundation for Innovation in Medicine of New Jersey, USA.

The Word nutraceutical is an amalgamation of the term **NUTRITION** and **PHARMACEUTICAL** or it can be more correctly defined as parts of a food that have a medical or health benefit including the prevention and treatment of disease.

The three main constituents, which make-up nutraceutical are **Herbal and related extracts, vitamins, minerals and nutrients**. Antioxidants and herbal teas also form an important part of the nutraceuticals market. The leading **antioxidant phytochemicals in demand are Vitamin A, C and E; carotenoids and flavonoids**.

Nutraceuticals are the most progressing sector for the health food and pharmaceutical industry based on plants. Many function food/nutraceutical companies are part of large food or pharmaceutical industries. A number of large food pharmaceutical companies, such as Abbott Laboratories, Himalayas, Dabur, Allen laboratories are also manufacturing nutraceuticals.^[12]

Classification of Nutraceuticals

In order to distinguish between the wide varieties of products there are multiple different types of products that fall under the category of nutraceuticals:

1) Dietary supplements:

- A Dietary supplement is a product that contains nutrients derived from food products that are concentrated in liquid or capsule form.
- Dietary supplements include – vitamins, minerals, co-enzyme Q, carnitine etc.,
- The Dietary Supplementation Health Education Act [DSHEA] formally defined “dietary supplement” using several criteria.^[14]

2) Functional Foods^[15]

3) Medical Foods^[16]

4) Pharmaceuticals^[17]

Nutraceutical Product Contain Calicum

- 1. CALCIUM CARBONATE^[18]**
- 2. VITAMIN D3 (Cholecalciferol)^[19]**
- 3. MAGNESIUM (Magnesium citrate)^[20]**
- 4. VITAMIN K2 (MK-7)^[21]**
- 5. ZINC (Zinc Picolinate)^[22]**
- 6. Boron (Boron citrate)^[23]**

Milk Powder

Powdered milk may not seem like the most appetizing ingredient, but it's actually part of what makes various breads and prepared foods delicious. Additionally, powdered milk is an important resource for people who don't have much access to fresh milk and can even help you get essential vitamins and nutrients in the wake of a disaster. There's a reason survivalists recommend stocking up on powdered milk — it's reliable and very well may be your only source of calcium, protein, or vitamins A and D after a crisis.

Yogurt

Yogurt is a dairy product obtained by fermentation of milk specific microorganisms, which shall be viable, active and abundant in the product. Yogurt is described by the United Nations' Food and Agriculture Organization and the World Health Organization in their Codex Alimentarius as a fermented milk product containing two strains of live

bacteria, *Lactobacillus delbrueckii* subsp. *bulgaricus* and *Streptococcus thermophilus*. Both strains must remain active in the final product, with a total of at least 10 million bacteria per gram.

Aim & Plan of Work

- Selection of pharmaceutical and nutraceutical product based on availability.
- To detect the calcium various selected pharmaceutical product & nutraceutical product by classical method and non-classical method.
 - Flame test
 - Solubility test
 - Visible spectrophotometric method
- To estimate the calcium various selected pharmaceutical and nutraceutical product by classical method and non-classical method
 - ✓ Complexometric titration
 - ✓ Acid base titration
 - ✓ Visible spectrophotometric method
- To compare the results, obtain from the general method for detection and estimation.

Qualitative Analysis Test

Identification Test for Calcium Lactate

Test	Experiment	Observation	Interference
A	Soluble in water, practically insoluble in ethanol	White precipitate is formed.	Presence of calcium lactate
B	Hold the spatula in the flame and observe the colour	Brick red or orange color is formed	Presence of calcium lactate

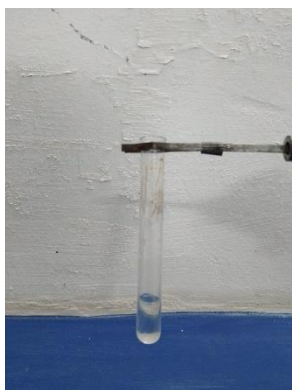


Fig.1: Test-A.



Fig. 2: Test-B.

Quantitative Analysis by UV-Spectroscopy

Preparation of Reagents and Standard Solutions

1. Preparation of **Calcium Stock Solution**: 0.03675 g of $\text{CaCl}_2 \cdot 2\text{H}_2\text{O}$ was weighed and diluted with distilled water to 100 mL, to obtain a concentration of 100 ppm. Then 1 mL of the solution was taken and diluted with distilled water to 100 mL to obtain a calcium standard of 1 ppm.
2. Preparation of **0.1 N NaOH Solution**: 0.1 N NaOH solution was made by weighing 0.4 grams of NaOH dissolved in distilled water to a volume of 100 ml.

3. Preparation of **0.5% murexide solution**: Weighed 50.0 mg of Murexide, and dissolved it in 10 mL of distilled water, to obtain a Murexide solution with a concentration of 0.5%. Add 25.0 mL of 96% ethanol to the murexide solution.

Assay of Calcium Lactate

- ❖ 1 ml of calcium lactate solution with a concentration of 1 ppm was taken.
- ❖ Put into a 50 ml measuring flask and 1.0 ml of murexide was added,
- ❖ 2.0 ml of 0.1 N NaOH (check pH 12-13), then distilled water was added until the volume was 50 ml.
- ❖ The solution was shaken until homogeneous, and then the absorbance was read at wavelength between 400 – 800 nm.^[26]



Fig. 3: Calcium Lactate Tablet

For Titration (Calcium Lactate and Calcium Gluconate)

Standardisation of Disodium EDTA

Weigh accurately about 0.3gm of anhydrous $MgSO_4$ and dissolve 50 ml of water. Add 10ml of ammonia buffer and 50mg mixture of mordant black II and sodium chloride (1:99) as indicator and titrate with disodium edetate until the solution becomes blue. Each ml of 0.05M disodium edetate is equivalent to 0.012325g of $MgSO_4$.^[27]

$$\begin{aligned} \text{MOLARITY OF DISODIUM EDTA} &= \frac{\text{Weight taken} \times \text{Expected molarity}}{\text{Titrate value} \times \text{Eq.factor}} \\ &= \frac{0.3 \times 0.05}{25 \times 0.12325} \\ &= 0.05M \end{aligned}$$

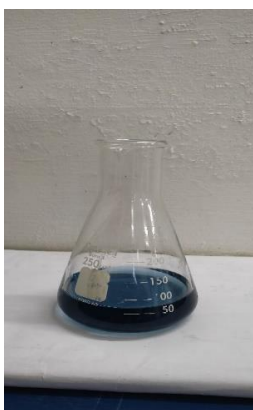


Fig. 4: Standardization of 0.5 EDTA.



Fig. 5: Standardization of 0.5M EDTA end point.

For Titration**Method of Assay**

Dissolve about 0.3g of previously dried sample, accurately weighed, in 50 ml of water. While stirring, add about 5 ml 0.05M of magnesium sulphate. Then add 10 ml of strong ammonia using 0.2g black mordant II indicator, and continue the titration to a blue end-point. Each ml of 0.05 M disodium ethylenediaminetetraacetate is equivalent to 0.01091 of $C_6H_{10}CaO_6$.

Percentage Purity of Calcium Lactate =

$$\frac{(\text{Titrate value} - 5) \times \text{Equivalent factor} \times \text{Eq. factor}}{\text{weight taken} \times \text{Expected molarity}} \times 100$$

$$= \frac{(27-5) \times 0.02242 \times 0.05}{0.5 \times 0.05} \times 100$$

$$= \frac{22 \times 0.02242 \times 0.05}{0.5 \times 0.05} \times 100$$

$$= 0.9864 \times 100$$

$$= \mathbf{98.64\%}$$



Fig. 6: Assay of Calcium lactate End Point.



Fig. 7: Assay of Calcium Lactate.

Qualitative analysis**Identification test for calcium gluconate**

Test	Experiment	Observation	Interence
A	Soluble in water, practically insoluble in ethanol	White precipitate is formed.	Presence of calcium gluconate
B	Hold the spatula in the flame and observe the colour	Brick red or orange color is formed	Presence of calcium gluconate



Fig. 8: TEST-A.



Fig. 9: TEST-B.

Quantitative Analysis for UV-Spectroscopy

Assay of Calcium Gluconate

- ❖ 1 ml of calcium gluconate solution with a concentration of 1 ppm was taken.
- ❖ Put into a 50 ml measuring flask and 1.0 ml of murexide was added,
- ❖ 2.0 ml of 0.1 N NaOH (check pH 12-13), then distilled water was added until the volume was 50 ml.
- ❖ The solution was shaken until homogeneous, and then the absorbance was read at wavelength.



Fig. 10: Calcium Gluconate Tablet.



Fig. 11: Detection of pH Calcium Lactate and Calcium Gluconate.

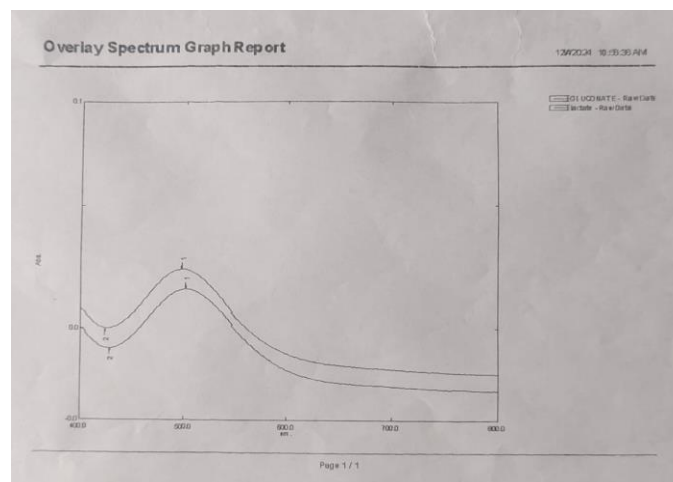


Fig. 12: Overlay Spectrum Graph Calcium Lactate and Calcium Gluconate.

For Titration

Method of Assay

Weigh accurately about 0.5gm sample dissolved in 50 ml of warm water, cool, while stirring, add about 5 ml 0.05M of magnesium sulphate. Then add 10 ml of strong ammonia using black mordant II indicator, and continue the titration to

a blue end-point. Each ml of 0.05M disodium ethylenediaminetetraacetate is equivalent to 0.02242g of $C_{12}H_{22}CaO_{14} \cdot H_2O$.^[28]

Percentage Purity of Calcium Gluconate =

$$\frac{(\text{Titrate value} - 5) \times \text{Equivalent factor} \times \text{Eq.factor}}{\text{weight taken} \times \text{Expected molarity}} \times 100$$

$$= \frac{(26-5) \times 0.02242 \times 0.05}{0.5 \times 0.05} \times 100$$

$$= \frac{21 \times 0.02242 \times 0.05}{0.5 \times 0.05} \times 100$$

$$= 0.96164 \times 100$$

$$= \mathbf{96.16\%}$$



Fig. 13: Assay of Calcium gluconate End Point.



Fig. 14: Assay of Calcium gluconate.

Qualitative Analysis

Milk Powder: (Dairy Whitener)

Into 2 test tubes,

- 1) 1 ml of the digested sample into test tube number 1, add a few drops of ammonia and there is no precipitate because the calcium hydroxide dissolves quite a lot. With precipitating agents that have been made for a long time, turbidity may occur due to the formation of calcium carbonate.
- 2) 1 ml of the digested sample into test tube number 2, add a few drops of dilute sulfuric acid and a white precipitate of calcium sulfate will form.^[29]



Fig. 15: Chemical Test for Milk Powder.

Quantitative Analysis by UV-Spectroscopy

Milk Powder

- ❖ 1 ml of dry digestion filtrate was taken and then neutralized with 0.1 N NaOH.
- ❖ The volume of the solution was made up to 10 ml with distilled water. 0.02 ml of the solution was taken and put into a 25 ml measuring flask.
- ❖ Then 1 ml of murexide solution and 2 ml of 0.1N NaOH were added.
- ❖ The volume of the solution is added with distilled water up to the limit mark.
- ❖ The solution is shaken until homogeneous then the absorbance is read at the maximum absorption wavelength.^[30]

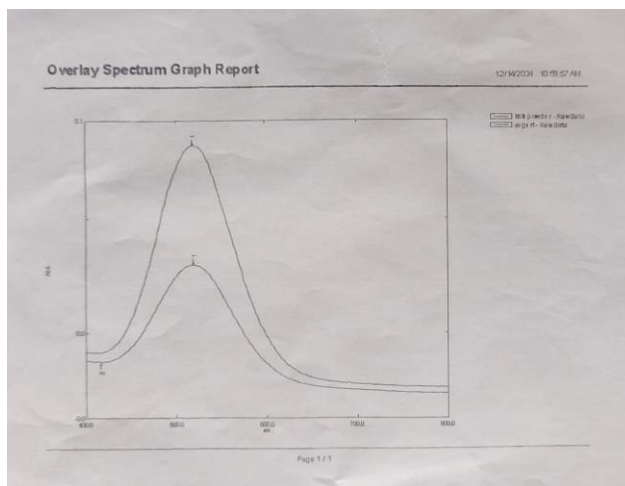


Fig. 16: Overlay Spectrum Graph Milk Powde.

For Titration

Procedure

- Prepare a 0.1 N NaOH solution by dissolving 4g of NaOH pellets in 1 L of distilled water.
- Weigh 1-2 g of milk powder and transfer it to a 100 ml Erlenmeyer flask.
- Add 20-30 ml of distilled water to the milk powder and mix well using a vortex mixer to create a uniform dispersion.
- Add 2-3 drops of phenolphthalein indicator to the milk powder dispersion. The indicator will turn pink if the milk powder is acidic.
- Fill the burette with the NaOH solution and record the initial reading. Slowly add the NaOH solution to the milk powder dispersion while stirring constantly. Continue adding the NaOH solution until the pink color disappears.
- Record the volume of NaOH solution added at the endpoint (when the pink color disappears).

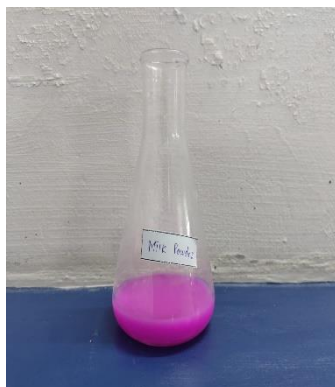
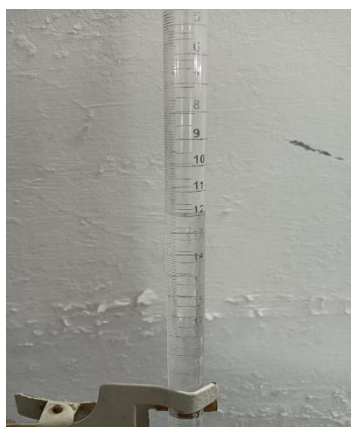
Formula

Acidity (%) = (Volume of NaOH solution added x Normality of NaOH solution x 100) / Weight of milk powder sample

- Initial burette reading: 0.00 ml
- Final burette reading: 12.00 ml
- Volume of NaOH solution added: 12.00 ml
- Normality of NaOH solution: 0.1 N
- Weight of milk powder sample: 1.5 g

Calculation

$$\begin{aligned}\text{Acidity (\%)} &= \frac{12.50 \times 0.1 \times 100}{1.5} \\ &= 9.33\%\end{aligned}$$

**Fig. 17: Assay of Milk Powder.****Fig. 18: Assay of Milk Powder End Point.****Qualitative Analysis****Yogurt: (Milky Mist)**

- ✓ A total of 1 ml. of sample is taken and poured into a test tube. Add 1 ml. 0.1 N hydrochloric acid and 1 ml. Na S.
- ✓ Samples containing calcium are indicated by the formation of a white precipitate.

**Fig. 19: Chemical Test for Yogurt.**

QUANTITATIVE ANALYSIS BY UV-SPECTROSCOPY

- ❖ The filtered yogurt drink sample (100 ml) was collected and transferred to a 50 ml volumetric flask.
- ❖ Then add 1 ml of murexide solution and 2 ml of 0.1 N sodium hydroxide solution.
- ❖ 50 ml of distilled water were added to the solution.
- ❖ A total of 1 ml of the sample solution was diluted with distilled water in a 25 mL volumetric flask.
- ❖ After being shaken until evenly distributed, the solution was transferred into a cuvette.
- ❖ At the highest possible wave, the absorbance was measured. The experiment was repeated twice⁽³¹⁾.

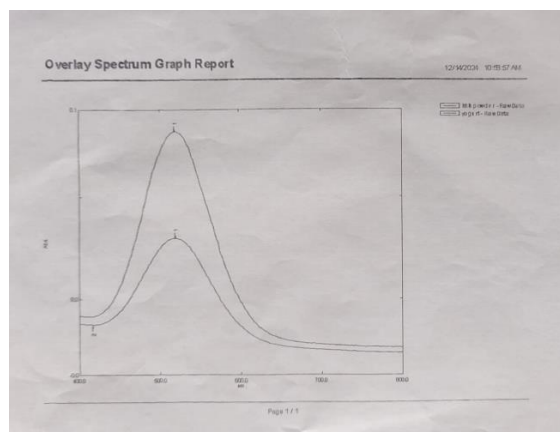


Fig. 20: Overlay Spectrum Graph Yogurt.

For Titration**Procedure**

- Prepare a 0.1 N NaOH solution by dissolving 4 g of NaOH pellets in 1 L of distilled water.
- Take a 10 mL sample of curd and transfer it to a 100 mL Erlenmeyer flask.
- Add 2-3 drops of phenolphthalein indicator to the curd sample. The indicator will turn pink if the curd is acidic.
- Fill the burette with the NaOH solution and record the initial reading. Slowly add the NaOH solution to the curd sample while stirring constantly. Continue adding the NaOH solution until the pink color disappears.
- Record the volume of NaOH solution added at the endpoint (when the pink color disappears).

Formula

Acidity (%) = (Volume of NaOH solution added x Normality of NaOH solution x 100) / Weight of milk powder sample

- Initial burette reading: 0.00 ml
- Final burette reading: 19.00 ml
- Volume of NaOH solution added: 19 ml
- Normality of NaOH solution: 0.1 N
- Weight of milk powder sample: 1.5 g

Calculation

$$\text{Acidity (\%)} = \frac{15.20 \times 0.1 \times 100}{10}$$

$$= 15.2 \%$$



Fig. 21: Assay of Yogurt.



Fig. 22: Assay of Yogurt End Point.

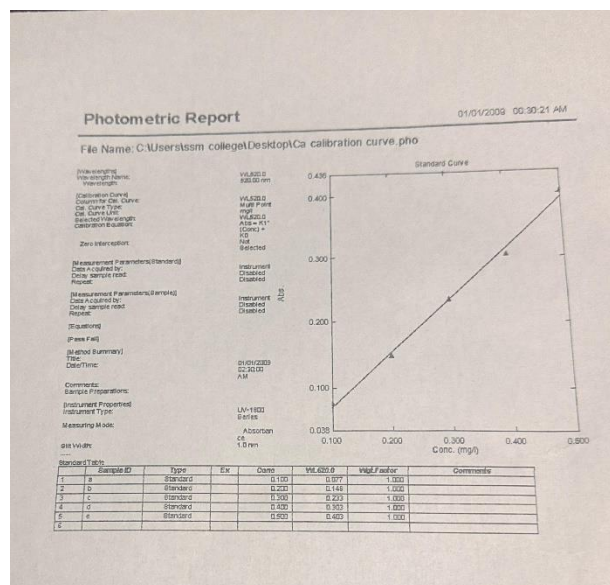


Fig. 23: Standard Calibration Curve.

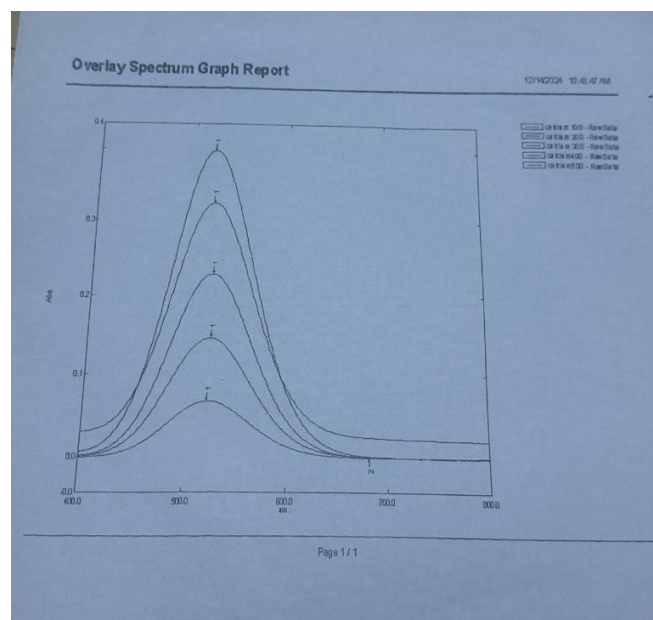


Fig. 24: Overlay of Calibration Curve.

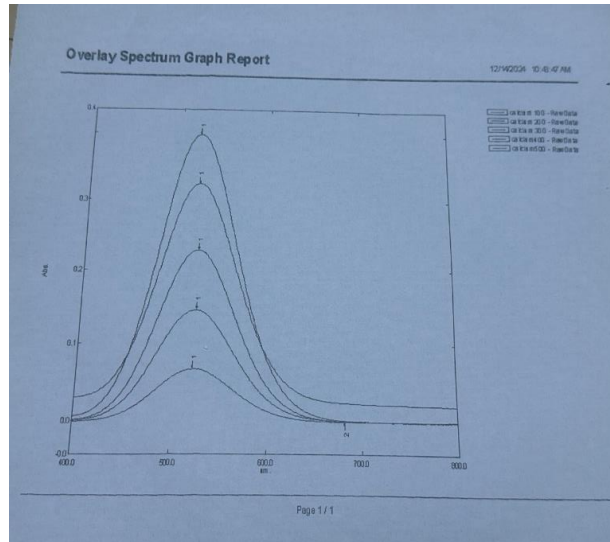


Fig. 25: Overlay of Calibration Curve.

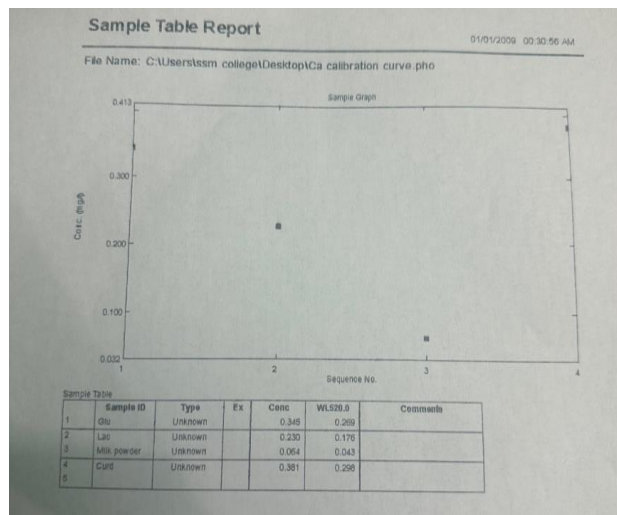


Fig. 26: Sample Calibration Curve.

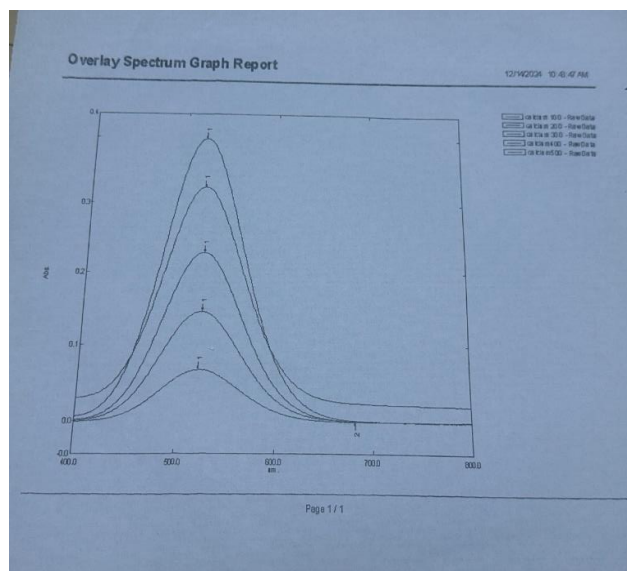


Fig. 27: Overlay of Calibration Curve.

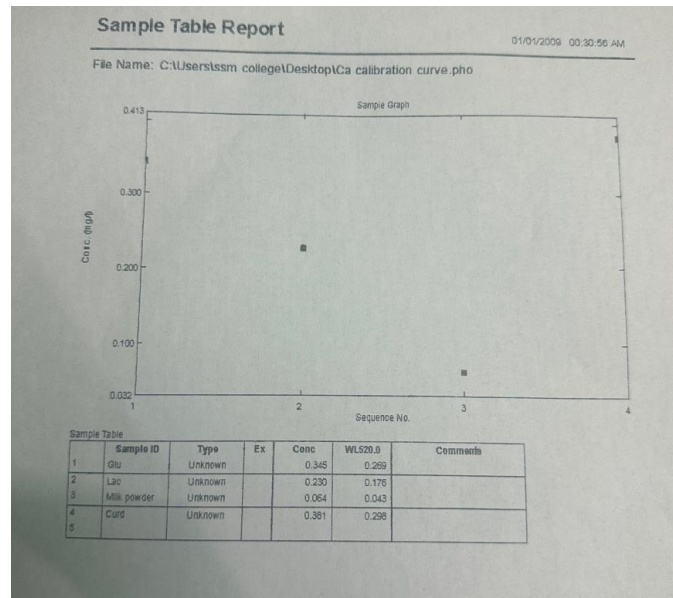


Fig. 28: Sample Calibration Curve.

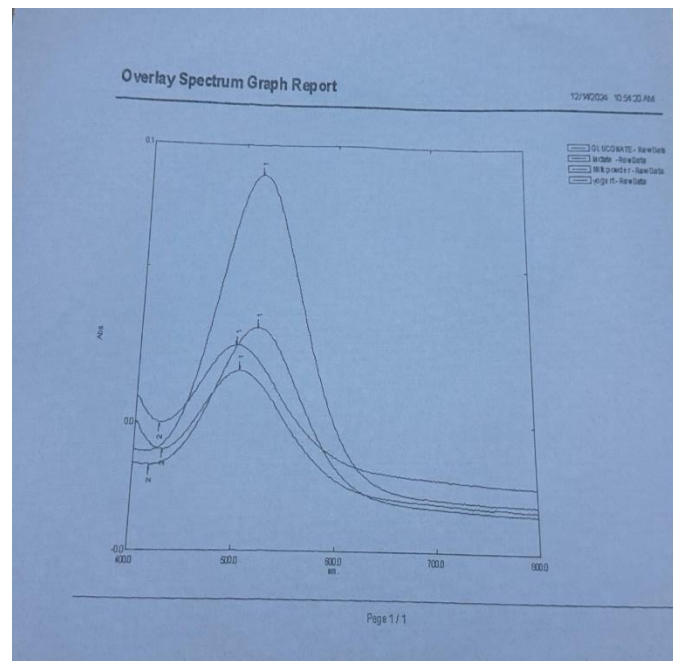


Fig. 29: Overlay of Sample Calibration Curve.

RESULT

Qualitative Test Results

Formulation	Chemical Test		Instrument λ Max
Calcium Lactate	+	+	520.1
Calcium Gluconate	+	+	520.4
Milk Powder	+	+	520
Yogurt	+	+	520.4

Quantitative Test Result

Formulation	UV Percentage	Titration	Limits
Calcium Gluconate	97.76%	96.41%	96 – 100%
Calcium Lactate	100.33%	98.64%	98-102%
Milk Powder	12%	9.33%	-
Yogurt	18.1%	15.2%	-

CONCLUSION

In this project, we compare the results of the calcium content in the two groups of the pharmaceutical product like calcium lactate IP 300mg & calcium gluconate 500 mg and other group of nutraceutical dietary supplement of milk powder and yogurt. For qualitative and quantitative analysis.

In qualitative analysis, a flame test was carried out and the colour of flame brick red colour which is classical method (non-instrumental method) and another non-classical method (instrumental method), Visible spectrophotometric method shown the λ_{max} at 520nm, which indicates the presence of calcium invariably in all selected pharmaceutical and nutraceutical product.

In quantitative analysis, a volumetric method of quantitative chemical analysis is in the classical method which is still widely used in the determination of calcium visible spectrophotometric (calorimetry) as method as quantitative analysis is in the non-classical method or instrumental method, which is tried in the determination of calcium there was no statically significant difference calculate and theoretical value in the estimation of calcium by volumetric method and visible spectrophotometric methods for all sample. As well as identification of calcium in all samples by flame test and λ_{max} determination also possible.

On that obtained result basis, it can be concluded that in classical method like flame test and volumetric analysis were carried out separately for the identification and estimation which have individual process, but in non-classical method like visible spectrophotometry method used simultaneously for the identification / estimation which have combined process of calcium in various selected samples.

So for the person who carried out the analysis of calcium regarding qualitatively by volumetric analysis is lengthier procedure and again flame test was carried out for qualitative purpose separately, but while visible spectrophotometric method used is short procedure and simultaneously qualitative and quantitative analysis were possibly by checking the λ_{max} at 520 and its absorbance.

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